

**REMARKS**

Claims 1-180 were pending in the present application. Claims 1-98, 100-152 and 156-180 are rejected, and claim 99 is objected to. Claims 153-155 have been withdrawn from consideration. By virtue of this response, claims 1, 4, 8, 10, 14-16, 18-19, 52, 58-59, 83-87, 90, 132-33, and 148-152 are amended, claims 7, 13, 17, 103-131, 147, and 156-180 are cancelled, and new claims 181-208 are added. Accordingly, claims 1-6, 8-12, 14-16, 18-102, 132-141, 148-152, and 181-208 are currently under consideration. Allowance of the pending claims is respectfully requested.

With respect to all amendments and cancelled claims, Applicant has not dedicated or abandoned any unclaimed subject matter and, moreover, has not acquiesced to any rejections and/or objections made by the Patent Office. Applicant reserves the right to pursue prosecution of any presently excluded claim embodiments in future continuation and/or divisional applications.

**Claim Amendments**

The amendments to claims 1, 4, 8, 10, 14-16, 18-19, 52, 58-59, 83-87, 90, 132-33, and 148-152, as well as new claims 181-208, are fully supported by the original application.

Claim 1 has been amended to indicate that the template DNA comprises a mixture of maternal DNA and fetal DNA and that the template DNA is obtained from a sample from a pregnant female. Support for these amendments can be found in original claims 7 and 17 (now cancelled). A minor change in verb tense has been made in claim 1 to more clearly indicate the claimed invention. In addition, the phrase "the presence or absence of" has been inserted in claim 1, per the suggestion made by the Office. Support for this amendment can be found at the end of claim 1.

Amendments have been made to claims 4, 8, 52, 59, 86, and 90 to eliminate certain members of the Markush groups recited therein. References to trademarks have been deleted from claim 52. Support for these amendments can be found in the corresponding original claims.

Claims 10, 58, 83, and 85 have been amended to correct typographical errors.

The claim dependencies have been corrected in claims 14-16 and 18-19 due to the amendment of claim 1 and cancellation of claims 13 and 17.

Claims 84 and 85 have been amended to indicate that the template DNA sequenced comprises a mixture of fetal DNA and maternal DNA and is obtained from a sample from a pregnant female. Support for these amendments can be found, e.g., in paragraphs [0155], [0157], and [0163]. (Paragraph numbers given herein for Applicant's application refer to the paragraph numbering in the Substitute Specification filed on May 23, 2005.)

The phrase "to form an annealed template and primer" has been inserted into step (3) of both claim 132 and claim 133 to provide proper antecedent basis. Support for this amendment can be found in step (4) of claim 132 and 133.

Claims 148-151 have been amended to correct the dependencies of these claims.

Claim 152 has been amended to indicate that the template DNA comprises a mixture of fetal DNA and maternal DNA and the template DNA is from a sample from a pregnant female. Support for this amendment can be found, e.g., in paragraphs [0155], [0157], and [0163] of the application and original claims 7 and 17.

New claims 181- 208 find support throughout the application as filed. For example, support for new claims 181-184 can be found in original claim 4. Support for new claims 185-188 can be found in original claim 8. New claim 189 finds support in original claim 11. New claim 190 finds support in paragraph [0155], new claim 191 finds support in paragraph [0153], and new claims 192-195 find support in paragraph [0047] of the application. New claim 196 finds support in original claim 63. New claims 197-200 find support, e.g., in original claim 86. New claim 201 finds support in original claim 96. Support for new claims 202-205 can be found, e.g., in paragraph [0154] of the application. New claim 206 finds support, e.g., in original claims 58 and 62. New claims 207 and 208 find support, e.g., in paragraphs [0139] and [0143], respectively, of the application.

No new matter is added by the amendments to the claims.

**Election/Restriction**

Applicant's previous election of Group I, claims 1-152 and 156-180, without traverse, is affirmed.

**Claim Rejections under 35 USC § 112 – 2<sup>nd</sup> Paragraph**

The Office has rejected claims 1-57, 103-142, 147 and 149-151 as being indefinite for failing to particularly point out and distinctly claim the subject matter which Applicant regards as the invention.

*Claim 1:* Claim 1 stands rejected as being indefinite for being unclear whether the phrase “wherein said heterozygous locus of interest was identified by determining the sequence of alleles at a locus of interest from template DNA” recites a required positive step. An *active* step of identifying the heterozygous locus of interest by determining the sequence of alleles at the locus of interest from template DNA is *not* a requirement of claim 1. However, claim 1 does require that the heterozygous locus be a heterozygous locus that has been identified by determining the sequence of alleles at a locus of interest from the indicated template DNA. Applicant submits that claim 1, as amended, is clear in this regard due to the verb tense in the phrase “*has been identified*” (emphasis added).

Claim 1 also stands rejected as being indefinite for failing to provide adequate nexus between the preamble and the claim steps. Claim 1 has now been amended to more clearly state in the preamble that the method is for detecting *the presence or absence of* a chromosomal abnormality.

Accordingly, Applicant respectfully requests that the rejection of claim 1 under 35 USC § 112, 2<sup>nd</sup> paragraph, be withdrawn.

*Claims 52, 103-131 and 147:* Claims 52, 103-131 and 147 stand rejected as indefinite because a trademark or trade name is included in the claim. The references to trademarks have been deleted from claim 52. Accordingly, Applicant respectfully requests that the rejection of claim 52

under 35 USC § 112, 2<sup>nd</sup> paragraph, be withdrawn. Claims 103-131 and 147 are cancelled, so the rejection of these claims under 35 USC § 112, 2<sup>nd</sup> paragraph, is moot.

*Claims 132-133:* Claims 132-133 are rejected as indefinite in that the phrase “annealed template and primer in step (4)” lacks proper antecedent basis. As amended, the claims no longer lack proper antecedent basis. Applicant respectfully requests that the rejection of claims 132-133 under 35 USC § 112, 2<sup>nd</sup> paragraph, be withdrawn.

*Claims 149-151:* Claims 149-151 stand rejected as indefinite for lacking proper antecedent basis. As amended claims 149-151 no longer lack proper antecedent basis. Applicant respectfully requests that the rejection of claims 149-151 under 35 USC § 112, 2<sup>nd</sup> paragraph, be withdrawn.

#### **Claims Rejections under 35 USC § 102**

*Claims 1-4, 7-8, 52, 56-57 and 152:* The Office has rejected claims 1-4, 7-8, 52, 56-57 and 152 as being anticipated by Antonarakis et al. (US 2005/0037388). Applicant respectfully traverses this rejection.

Claim 1, as amended, is directed to a method for detecting the presence or absence of a fetal chromosomal abnormality, comprising quantitating the relative amount of the alleles at a heterozygous locus of interest, wherein said heterozygous locus of interest has been identified by determining the sequence of the alleles at a locus of interest from template DNA obtained from a sample from a pregnant female, wherein the relative amount is expressed as a ratio, wherein said ratio indicates the presence or absence of a fetal chromosomal abnormality, and wherein the template DNA comprises a mixture of maternal DNA and fetal DNA. Claims 2-4, 7-8, 52, 56-57 all depend (directly or indirectly) from independent claim 1. Claim 152, as amended, is directed to a method for detecting the presence or absence of a fetal chromosomal abnormality, comprising both (a) determining the sequence of alleles of a locus of interest from template DNA, wherein the template DNA comprises a mixture of fetal DNA and maternal DNA, and wherein the template DNA is from a sample from a pregnant female, and (b) quantitating the relative amount of the alleles at a heterozygous locus of interest that was identified from the locus of interest of (a),

wherein said relative amount is expressed as a ratio, and wherein said ratio indicates the presence or absence of a fetal chromosomal abnormality.

Examples 14 and 18-23 of Applicant's specification provide some non-limiting, specific examples of the methods of claims 1 and 152. Paragraphs [0983]-[0987] of Example 14, for instance, provide a description of how fetal chromosomal abnormalities can be determined by analyzing SNPs using template DNA from plasma isolated from the blood of a pregnant female, even though that template DNA comprises a mixture of free fetal DNA and free maternal DNA. Example 14 further describes a study in which a SNP analysis is performed on samples comprising mixtures of DNA from children afflicted with Down's syndrome and maternal DNA in various proportions designed to simulate the mixture of fetal DNA and maternal DNA that would be expected to be isolated from the plasma samples of pregnant women carrying fetuses that are afflicted with Down's syndrome (paragraphs [0988] –[1034]). The presence of the Trisomy 21 condition was identified in numerous samples that contained various percentages of abnormal DNA, such as a sample containing 40% Down syndrome DNA and 60% maternal DNA, using the claimed methods (Table XX and paragraphs [1015]-[1032]). In addition to the simulated study described in Example 14, descriptions of examples of methods of detecting fetal chromosome abnormalities by performing SNP analysis on mixtures of fetal DNA and maternal DNA are provided in Examples 18-23 of Applicant's specification.

To anticipate a claim, a prior art reference must teach or suggest each and every limitation of the claim. Applicant respectfully submits that Antonarakis et al. does not anticipate claims 1-4, 7-8, 52, 56-57 and 152, because the reference fails to disclose or suggest all elements of claims 1-4, 7-8, 52, 56-57 and 152. By contrast to the methods of claims 1-4, 7-8, 52, 56-57 and 152, which all involve quantitating the relative amount of *alleles* at *a* locus of interest, the methods for detecting chromosomal abnormalities taught by Antonarakis et al. involve the quantification and comparison of the relative amounts of paralogous sequences at *different* chromosomal locations, such as paralogous genes (i.e., "genes that have a common evolutionary origin but which have been duplicated over time in the human genome" (paragraph [0076] of Antonarakis et al.)). See, e.g., abstract and paragraphs [0011] and [0073] of Antonarakis et al. The paralogous sequences used in

the methods taught by Antonarakis are preferably on different chromosomes, although they may also be on different arms of a chromosomal (see, e.g., paragraphs [0011] and [0073] of Antonarakis et al.). Nowhere does Antonarakis teach or suggest measuring the relative amounts of sequences found at the *same* locus. Nowhere does Antonarakis et al. teach or suggest that *alleles* at a particular locus be sequenced and/or that the relative amounts of *alleles* for a particular heterozygous locus be determined.

In addition, claims 1-4, 7-8, 52, 56-57 and 152, as amended, differ from the methods taught in Antonarakis et al. in that the template DNA in the methods of the claims comprise a *mixture* of fetal DNA and maternal DNA, whereas Antonarakis et al. does not teach or suggest the use of a mixture of fetal DNA and maternal DNA as template DNA for analysis. Rather, Antonarakis et al. teaches the isolation of fetal DNA from isolated fetal cells as the source of nucleic acids for analysis. See, e.g., claim 1, paragraphs [0098]-[0100], [0139], and [0144] of Antonarakis et al.

Since Antonarakis et al. does not teach or suggest each and every element of claims 1-4, 7-8, 52, 56-57 and 152, Applicant respectfully requests that the rejection of claims 1-4, 7-8, 52, 56-57 and 152 under 35 USC § 102 be withdrawn.

Claims 87-97, 100 and 102: The Office has rejected claims 87-95, 100 and 102 under 35 USC § 102(b) as being anticipated by Lo et al. (WO98/39474). Applicant respectfully traverses this rejection.

Claim 87 is directed to a method for preparing a sample for analysis comprising isolating free nucleic acid from a sample that contains nucleic acid, wherein an agent that inhibits cell lysis has been added to the sample to inhibit lysis of cells, if cells are present, wherein the agent is selected from the group consisting of membrane stabilizer, cross-linker, and cell lysis inhibitor. Claims 88-97, 100, and 102 all depend (directly or indirectly) from claim 87.

As indicated in the application, Applicant has discovered that the addition of a cell lysis inhibitor during the sample preparation process can significantly and unexpectedly increase the proportion of fetal DNA versus maternal DNA obtained from a sample such as a plasma sample

obtained from the blood of a pregnant woman. Examples of the impact of using a cell lysis inhibitor such as formalin to inhibit cell lysis during the sample preparation process are provided in Example 4 of Applicant's specification. As shown in Example 4, the amounts of fetal DNA isolated from maternal blood samples were significantly, and unexpectedly, higher for samples treated with both formalin and EDTA versus those treated with EDTA alone. As indicated in paragraph [0490] in Example 4, in one set of experiments, the percentage of fetal DNA present in the sample treated with only EDTA was found to be 1.56%, whereas the percentage of fetal DNA in the sample treated with both formalin and EDTA was found to be 25%. In another set of experiments, the lower and upper range of the mean percentage of fetal DNA isolated from samples treated with both formalin and EDTA were found to be 19.47% and 43.69%, whereas the lower and upper range of fetal DNA isolated from the same samples with EDTA only were found to be 7.71% and 22.1% (paragraph [0491] and Table V of Example 4). The high percentage of fetal DNA versus maternal DNA which can be obtained from the plasma of maternal blood to which cell lysis inhibitor (e.g., formalin) has been added is further demonstrated in Example 15 of Applicant's specification (see, e.g., Table XXI).

As already noted, a prior art reference must teach or suggest each and every limitation of the claim. Applicant respectfully submits that Lo et al. does not anticipate claims 87-95, 100 and 102, since Lo et al. does not teach or suggest each and every element of the claims. Lo et al. describes the collection of blood into a tube containing EDTA and the Office has asserted that EDTA is an agent that inhibits cell lysis. However, the Office has provided absolutely no documentary evidence or rationale in support of its assertion that EDTA is an agent that inhibits cell lysis. In effect, the Office has taken official notice regarding the nature of EDTA without providing the required supporting documentary evidence. Applicant asserts that the (alleged) functioning of EDTA as a cell lysis inhibitor is not "capable of instant and unquestionable demonstration as being well-known" and therefore must be supported by evidentiary support and/or an adequate technical line of reasoning (MPEP 2144.03) Accordingly, Applicant respectfully requests that proper documentary evidence or an adequate technical line of reasoning required under MPEP 2144.03 be provided in support of the Office's assertion regarding EDTA if the rejection is to be maintained.

In addition to being improperly supported by documentary evidence, the assertion by the Office that EDTA is a cell lysis inhibitor is simply incorrect. Applicant asserts that EDTA is not an “agent that inhibits cell lysis.” Rather, EDTA is a well-known chelator of calcium and magnesium. EDTA is routinely added to blood during the blood collection process as an anticoagulant due to its ability to chelate calcium. In fact, EDTA is sometimes included as an ingredient in cell lysis buffers. See, e.g., the cell lysis buffer described at column 7, lines 28-33, of U.S. Patent No. 6,100,029 (Lapidus et al.), which comprises 20 mM EDTA. EDTA is clearly referred to as a chelator in Applicant’s specification, not as a cell lysis inhibitor (see, e.g., paragraph [0165] of Applicant’s specification), whereas multiple examples of agents that inhibit cell lysis are provided separately (see, e.g., paragraphs [0166] to [0167]). As shown in Example 4, discussed above, the addition of formalin, even in the presence of EDTA, to samples has a dramatic effect on the percentage of free fetal DNA isolated from the samples. The fact that the addition of formalin can have such a dramatic effect on the percentage of free fetal DNA serves to demonstrate that formalin and EDTA have very different properties and cannot be equated to each other.

Since Lo et al. does not teach or suggest the use of an agent that inhibits cell lysis, Lo et al. does not teach and every element of claims 87-97, 100 and 102, and since the Office’s assertion that EDTA is a cell lysis inhibitor is wholly unsupported by documentary evidence or reasoning, the rejection of claims 87-97, 100, and 102 under 35 USC § 102 is improper. Applicant respectfully requests that the rejection of claims 87-97, 100 and 102 under 35 USC § 102 be withdrawn.

Claims 87-97, 100 and 102: The Office has also rejected claims 87-97, 100, and 102 as being anticipated by Schueler et al. (US2004/0185495). Applicant respectfully traverses this rejection.

As indicated above, claim 87 is directed to a method for preparing a sample for analysis comprising *isolating free nucleic acid* from a sample that contains nucleic acid, wherein an agent that inhibits cell lysis has been added to the sample to inhibit lysis of cells, if cells are present, and wherein said agent is selected from the group consisting of membrane stabilizer, cross-linker, and

lysis inhibitor. Claims 88-97, 100, and 102 all depend (directly or indirectly) from independent claim 87.

As previously stated, a prior art reference must teach or suggest each and every limitation of the claim. Applicant submits that Schueler et al. fails to teach or suggest every element of claims 87-97, 100, and 102. Schueler et al. is directed to methods for “identifying and diagnosing rare fetal cells in a mixed cell population such as a maternal blood sample”(Abstract of Schueler et al.) Schueler et al. teaches the use of fixatives, such as formaldehyde, to fix cells as part of methods designed to detect fetal cells within mixed cell populations, separate the cells, and/or analyze the cells in situ. See, e.g., paragraphs [0064] and [0223]-[0246] of Schueler et al. The reference teaches identification and analysis of fetal cells using nucleic acid probes that selectively target fetal nucleic acid (e.g., fluorescent in situ hybridization (FISH)). The Schueler et al. methods do not involve the *isolation* of free nucleic acid from samples containing nucleic acid, and therefore do not teach any preparation methods for such methods, let alone the use of fixatives like formalin in such methods.

Furthermore, the teachings of Schueler et al. that fixatives like formalin may be used in the preservation of intact fetal cells in no way would have suggested to one of ordinary skill in the art that the addition of an agent that inhibits cell lysis to samples would have provided any advantage in methods of isolating free fetal DNA. Absent Applicant’s teachings (e.g., Example 4), it would not have been apparent to one of ordinary skill in the art wishing to isolate free fetal DNA what advantage adding an agent used in cell preservation methods to the sample from which fetal DNA was to be isolated would have offered, and therefore, one of ordinary skill in the art would not have been motivated to do so. And it certainly would not have been obvious, absent Applicant’s teachings, that the addition of such an agent as formalin would increase the percentage of free fetal DNA isolated from the sample significantly (e.g., by 2-fold or more, as shown in Example 4 of Applicant’s specification).

Since Schueler et al. does not teach or suggest each and every element of claims 87-97, 100 and 102, Applicant respectfully requests that the rejection of claims 87-97, 100 and 102 under 35 USC § 102 be withdrawn.

Claims 156-180: The Office has rejected claims 156-180 as being anticipated by Jones et al. (US2003/0082576).

Without acquiescing to the arguments made by the Office regarding claims 156-180, and for the sole purpose of expediting prosecution, claims 156-180 are cancelled. Since claims 156-180 are cancelled, the rejection of claims 156-180 under 35 USC § 102 is moot.

### **Claim Rejections under 35 USC § 103**

Claims 5-6: The Office has rejected claims 5-6 as being unpatentable over Antonarakis et al. (US2005/0037388). Applicant respectfully traverses this rejection.

Claims 5 and 6 depend from independent claim 1, discussed above. In the methods of claims 5 and 6, alleles of multiple loci of interest are sequenced and their relative amounts quantitated and expressed as a ratio.

To establish a prima facie case of obviousness, the prior art references must teach or suggest each and every claim limitations. Applicant submits that the Office has not met its burden of establishing a proper prima facie case of obviousness, since Antonarakis et al. does not teach or suggest all elements of claims 5 and 6. As discussed above with respect to the rejection of claim 1 under § 102, Antonarakis et al. is directed to the analysis of paralogous sequences at different chromosomal locations, and does not teach or suggest the quantitation of the relative amount of *alleles* at a heterozygous locus of interest. Furthermore, Antonarakis et al. does not teach or suggest the use of a mixture of fetal and maternal DNA as template DNA.

Since Antonarakis et al. does not teach or suggest each and every element of claims 5-6, Applicant respectfully requests that the rejection of claims 5-6 under 35 USC § 103 be withdrawn.

Claims 9-19: The Office has rejected claims 9-19 as being unpatentable over Antonarakis et al. (US2005/0037388), as applied against claim 4, and further in view of Schueler et al. (US2004/0185495) or Lo et al. (WO98/39474). Applicant respectfully traverses this rejection.

Claims 9-19 depend (directly or indirectly) from independent claim 1 discussed above. These claims further require that the sample is mixed with an agent that inhibits cell lysis, wherein the agent is selected from the group consisting of membrane stabilizer, cross-linker, and cell lysis inhibitor.

As already noted, to establish a prima facie case of obviousness, the prior art references must teach or suggest each and every claim limitations. Applicant submits that Antonarakis et al. as applied against claim 4, and further in view of Schueler et al. or Lo et al., does not teach or suggest a method comprising all of the elements of claims 9-19. As discussed above with respect to the rejection of claim 1 under § 102, Antonarakis et al. does not teach or suggest methods comprising quantitating the relative amount of alleles at a heterozygous locus of interest nor the use of a mixture of fetal and maternal DNA as template DNA. Furthermore, the combination of Antonarakis et al. with Lo et al. does not teach or suggest all elements of claims 9-19 because, as discussed above regarding the rejections of claim 87 under § 102, Lo et al. does not teach that an agent that inhibits cell lysis is mixed with the sample.

Since Antonarakis et al. as applied against claim 4, and further in view of Schueler et al. or Lo et al., does not teach or suggest each and every element of claims 9-19, Applicant respectfully requests that the rejection of claims 9-19 under 35 USC § 103 be withdrawn.

Claims 58-62 and 64-83: The Office has rejected claims 58-62 and 64-83 as being unpatentable over Lo et al. (WO 98/39474) in view of Wallace et al. (US 5,639,611) or Jones et al. (US 2003/0082576). Applicant respectfully traverses this rejection.

Claim 58 is directed to a method for determining the sequence of a locus of interest on free fetal DNA from a sample comprising free fetal DNA wherein an agent that inhibits cell lysis has been added to the sample to inhibit the lysis of cells, if cells are present, wherein the agent is a membrane stabilizer, cross-linker, or cell lysis inhibitor. Claims 59-62 and 64-83 depend from independent claim 58.

As stated above, to establish a prima facie case of obviousness, the prior art references must teach or suggest each and every claim limitations. Applicant submits that Lo et al., in view of Wallace et al. or Jones et al., does not teach or suggest every element of claims 58-62 and 64-83, since as discussed above regarding the rejection of claim 87 over Lo et al. under § 102, Lo et al. does not teach or suggest the addition of an agent that inhibits cell lysis to a sample. As discussed above, the Office's assertion that EDTA is a cell lysis inhibitor is both improper for lack of support and incorrect.

Since Lo et al., in view of Wallace et al. or Jones et al., does not teach or suggest each and every element of claims 58-62 and 64-83, Applicant respectfully requests that the rejection of claims 58-62 and 64-83 under 35 USC § 103 be withdrawn.

Claim 63: The Office has rejected claim 63 as being unpatentable over Lo et al. (WO98/39474) in view of Wallace et al. (US 5,639,611) or Jones et al. (US 2003/0082576) as applied against claims 58 and 62 above and further in view of Schueler et al. (US 2004/0185495). Applicant respectfully traverses this rejection.

Claim 63 is directed to a method comprising determining the sequence of a locus of interest on free fetal DNA from a sample comprising free fetal DNA, wherein an agent that inhibits cell lysis has been added to the sample to inhibit lysis of cells, if cells are present, wherein the agent is a cell lysis inhibitor selected from the group consisting of glutaraldehyde, derivatives of glutaraldehyde, formaldehyde, derivatives of formaldehyde, and formalin.

To establish a prima facie case of obviousness, there must be some suggestion or motivation to modify the reference or to combine reference teachings. Applicant respectfully submits that there is no motivation to combine the teachings of the reference Schueler et al. with the teachings of the references Lo et al., Wallace et al., and Jones et al. As previously noted, the methods of Schueler et al. are directed to methods of detecting fetal cells and the analysis of fetal DNA *in situ*. Therefore, one of ordinary skill in the art would not be motivated to use the techniques used in such methods as part of a method involving the analysis of free fetal DNA as claimed in claim 63. Also, although the Office has suggested that the substitution of the cell isolation method taught by Lo et al. in view

of Wallace et al. or Jones et al. with the cell isolation method taught by Schueler et al. would be an obvious substitution, Applicant submits that, as discussed above with respect to the rejection of claim 87 under §102, EDTA simply cannot be equated with formalin, either in function or effect. Applicant submits that one of ordinary skill in the art would find nothing obvious about substituting a sample that has been treated with an anticoagulant, the known purpose of the EDTA added to blood in Lo et al., with a sample that has been treated with a method of fixing cells, the known purpose of Schueler et al.'s formaldehyde agents (see, e.g., paragraph [0229] of Schueler et al.). Furthermore, one of ordinary skill in the art would not have been motivated to combine a technique or reagent used in cell preservation and isolation methods, such as those taught by Schueler et al., with methods of analyzing fetal nucleic acids in plasma and serum, such as those taught by Lo et al. Contrary to the assertions of the Office, there is no "common known purpose" that would motivate one to combine the teachings of the cited references.

Since there is no motivation to combine Lo et al., Wallace et al. or Jones et al., and Schueler et al., Applicant respectfully requests that the rejection of claim 63 under 35 USC § 103 be withdrawn.

Claim 101: The Office has rejected claim 101 as being unpatentable over Lo et al. (W098/39474) or Schueler et al. (US 2004/0185495). Applicant respectfully traverses this rejection.

Claim 101 depends from independent claim 87, and, accordingly, is directed to a method for preparing a sample for analysis comprising isolating free nucleic acid from a sample that contains nucleic acid, wherein an agent that inhibits cell lysis has been added to the sample to inhibit lysis of cells, if cells are present, and wherein the agent is selected from the group consisting of membrane stabilizer, cross-linker, and cell lysis inhibitor. Claim 101 further comprises a centrifugation step, wherein the centrifugation step is performed with the centrifuge braking power set to zero.

As previously noted, to establish a prima facie case of obviousness, the prior art references much teach or suggest all claim limitations. Applicant respectfully submits that neither Lo et al. nor Schueler et al. teaches or suggests all elements of claim 101. As discussed above with respect to

the rejection of claim 87 over Lo et al., Lo et al. does not teach or suggest the use of a sample to which an agent that inhibits cell lysis has been added. Although the Office has asserted that EDTA is a cell lysis inhibitor, this is not correct and the assertion has been improperly made with no evidentiary support. Furthermore, as discussed above with respect to the rejection of claim 87 over Schueler et al., Schueler et al. fails to teach or suggest a method comprising isolating free nucleic acid from a sample, wherein a cell lysis inhibitor has been added to the sample to inhibit the lysis of cells.

Since neither Lo et al. nor Schueler et al. teaches or suggests each and every element of claim 101, Applicant respectfully submits that the rejection of claim 101 under 35 USC §103 be withdrawn.

Claims 156-180: The Office has rejected claims 156-180 as being unpatentable over Shapero et al. (Genome Research 11: 1926-1934) in view of the Stratagene Catalog (1988).

Without acquiescing to the arguments made by the Office regarding claims 156-180, and for the sole purpose of expediting prosecution, claims 156-180 are cancelled. Since claims 156-180 are cancelled, the rejection of claims 156-180 under 35 USC § 103 is moot.

Claims 1-8 and 152: The Office has rejected claims 1-8 and 152 as being unpatentable over Lapidus et al. (US 6,100,029). Applicant respectfully traverses this rejection.

As indicated above, claim 1, as amended, is directed to a method for detecting the presence or absence of a fetal chromosomal abnormality, comprising quantitating the relative amount of the alleles at a heterozygous locus of interest, wherein said heterozygous locus of interest has been identified by determining the sequence of alleles at a locus of interest from template DNA obtained from a sample from a pregnant female, wherein the relative amount is expressed as a ratio, wherein the ratio indicates the presence or absence of a fetal chromosomal abnormality, and wherein the template DNA comprises a mixture of maternal DNA and fetal DNA. Claims 2-8 depend from independent claim 1. Claim 152, as amended, is also directed to a method for detecting the presence or absence of a fetal chromosomal abnormality, comprising (a) determining the sequence

of alleles of a locus of interest from template DNA, wherein the template DNA comprises a mixture of fetal DNA and maternal DNA, and wherein the template DNA is from a sample from a pregnant female, (b) quantitating the relative amount of the alleles at a heterozygous locus of interest that was identified from the locus of interest of (a), wherein said relative amount is expressed as a ratio, and wherein said ratio indicates the presence or absence of a fetal chromosomal abnormality.

As already noted, to establish a prima facie case of obviousness, the prior art references must teach or suggest all claim limitations. Applicant submits that Lapidus et al. fails to teach or suggest all claim limitations of claims 1-8 and 152. Although Lapidus et al. teaches the analysis of fetal DNA, Lapidus et al. does not teach or suggest methods involving the quantitation and expression as a ratio of the relative amount of alleles from template DNA comprising a mixture of fetal DNA and maternal DNA. Rather, Lapidus et al. appears to teach methods involving the quantitation and comparison of amounts of the maternal allele found in fetal DNA versus the amounts of the paternal allele found in the fetal DNA (see, e.g., line 64, column 2, to line 3, column 3, and lines 40-43, column 4, of Lapidus et al.), hence the expectation that “a statistically-significant difference between the two amounts is indicative of an aneuploidy or mutation in the chromosome.” Nowhere does Lapidus et al. teach the quantitation of the relative amounts of one allele in template DNA comprising a mixture of fetal DNA and maternal DNA versus another allele in the template DNA comprising the mixture. The mere observance of a statistically-significant difference between the amounts of the two alleles in such a mixture would not necessarily be indicative of a chromosomal abnormality, where, for instance, the fetus is heterozygous and the mother homozygous. Lapidus et al. also teaches the enumeration of fetal chromosomes (see, e.g., column 2, lines 27-31), but not the enumeration of chromosomes that represent a mixture of fetal and maternal chromosomes. Since the methods of Lapidus et al. are directed toward analyzing fetal DNA, rather than a mixture of fetal DNA and maternal DNA, Lapidus et al. naturally teaches the isolation of fetal DNA from fetal cells (e.g., line 65, column 6, to line 37, column 7.)

Since Lapidus et al. does not teach or suggest each and every element of claims 1-8 and 152, Applicant respectfully requests that the rejection of claims 1-8 and 152 under 35 USC § 103 be withdrawn.

Claims 9-19: The Office has rejected claims 9-19 as being unpatentable over Lapidus et al. (US 6,100,029) as applied against Claim 4 above and further in view of Schueler et al. (US 2004/0185495) or Lo et al. (WO98/39474). Applicant respectfully traverses this rejection.

Claims 9-19 all depend from claim 1, as amended, and therefore incorporate all elements of the method of claim 1 as described above. In addition, in the methods of each of the claims 9-19, the sample is mixed with an agent that inhibits cell lysis, wherein the agent is selected from the group consisting of membrane stabilizer, cross-linker, and cell lysis inhibitor.

Again, to establish a prima facie case of obviousness, the prior art references must teach or suggest all the claim limitations. Applicant submits that Lapidus et al. fails to teach or suggest all elements of claims 9-19. As discussed above, Lapidus et al. does not teach or suggest methods for detecting a fetal chromosomal abnormality, wherein the template DNA used in the analysis comprises a mixture of fetal DNA and maternal DNA. Furthermore, as also discussed above, Applicant submits that contrary to the unsupported assertions made by the Office, Lo et al. does not teach or suggest the use of a sample that has been mixed with an agent that inhibits cell lysis, since EDTA is not a cell lysis inhibitor.

Since Lapidus et al. in view of Schueler et al. or Lo et al. does not teach or suggest each and every element of claims 9-19, Applicant respectfully requests that the rejection of claims 9-19 under 35 USC § 103 be withdrawn.

Claims 20-21, 23-24, 27-30, 33-34, 37-39, 41, 44-52 and 56-57: The Office has rejected claims 20-21, 23-24, 27-30, 33-34, 37-39, 41, 44-52 and 56-57 as being unpatentable over Lapidus et al. (US 6,100,029) as applied against Claim 1 above and further in view of Jones et al. (US2003/0082576). Applicant respectfully traverses this rejection.

Claims 20-21, 23-24, 27-30, 33-34, 37-39, 41, 44-52 and 56-57 all depend (directly or indirectly) from independent claim 1 and therefore incorporate all elements of claim 1, as amended. Accordingly, claims 20-21, 23-24, 27-30, 33-34, 37-39, 41, 44-52 and 56-57 are all generally

directed to methods for detecting the absence or presence of a fetal chromosomal abnormality, in which the template DNA comprises a mixture of fetal DNA and maternal DNA.

As already noted, to establish a prima facie case of obviousness, the prior art references must teach or suggest all claim limitations. Applicant respectfully submits that Lapidus et al., alone or in combination with Jones et al., does not teach or suggest all elements of claims 20-21, 23-24, 27-30, 33-34, 37-39, 41, 44-52 and 56-57. For instance, Lapidus et al., alone or in combination with Jones et al., does not teach or suggest the use of template DNA comprising a mixture of fetal DNA and maternal DNA, as discussed above with respect to the rejection of claim 1 over Lapidus et al. under § 103.

Since Lapidus et al. in view of Jones et al. does not teach or suggest each and every element of claims 20-21, 23-24, 27-30, 33-34, 37-39, 41, 44-52 and 56-57, Applicant respectfully requests that the rejection of claims 20-21, 23-24, 27-30, 33-34, 37-39, 41, 44-52 and 56-57 under 35 USC § 103 be withdrawn.

Claim 22: The Office has rejected claim 22 as being unpatentable over Lapidus et al. (US 6,100,029) in view of Jones et al. (US2003/0082576) as applied against Claim 20-21 above and further in view of Western et al. (US 5,882,857). Applicant respectfully traverses this rejection.

Claim 22 depends from independent claim 1, as amended. Accordingly, claim 22 is directed to a method for detecting a chromosomal abnormality, in which the template DNA comprises a mixture of fetal DNA and maternal DNA.

To establish a prima facie case of obviousness, the prior art references must teach or suggest all the claim limitations. Applicant respectfully submits that Lapidus et al., alone or in combination with Jones et al. and/or Western et al., does not teach or suggest all elements of claim 22. Lapidus et al., alone or in combination with Jones et al. and/or Western et al., does not teach or suggest the use of template DNA comprising a mixture of fetal DNA and maternal DNA. (See the discussion above with respect to the rejection of claim 1 over Lapidus et al. under § 103.)

Since Lapidus et al. in view of Jones et al. and further in view of Western et al. does not teach or suggest each and every element of claim 22, Applicant respectfully requests that the rejection of claim 22 under 35 USC § 103 be withdrawn.

Claim 43: The Office has rejected claim 43 as being unpatentable over Lapidus et al. (US 6,100,029) in view of Jones et al. (US2003/0082576) as applied against Claim 20 and 21 above and further in view of MacLeod et al. (US 6,221,600). Applicant respectfully traverses this rejection.

Claim 43 depends from claim 1, as amended. Accordingly, claim 43 is directed to a method for detecting a chromosomal abnormality, in which the template DNA comprises a mixture of fetal DNA and maternal DNA.

To establish a prima facie case of obviousness, the prior art references must teach or suggest all the claim limitations. Applicant respectfully submits that Lapidus et al., alone or in combination with Jones et al. and/or MacLeod et al., does not teach or suggest all elements of claim 43. Lapidus et al., alone or in combination with Jones et al. and/or MacLeod et al., does not teach or suggest the use of template DNA comprising a mixture of fetal DNA and maternal DNA, as discussed above with respect to the rejection of claim 1 over Lapidus et al. under § 103.

Since Lapidus et al. in view of Jones et al. and further in view of MacLeod et al. does not teach or suggest each and every element of claim 43, Applicant respectfully requests that the rejection of claim 43 under 35 USC § 103 be withdrawn.

Claims 84-86: The Office has rejected claims 84-86 as being unpatentable over Jones et al. (US2003/0082576) in view of Lo et al. (W098/39474). Applicant respectfully traverses this rejection.

Claim 84, as amended, is directed to a method for determining the sequence of a locus of interest from template DNA, wherein the template DNA comprises a mixture of fetal DNA and maternal DNA and is obtained from a sample from a pregnant female. Claim 85, as amended, is directed to a method for determining the sequence of alleles of a locus of interest from template DNA, wherein the template DNA comprises a mixture of fetal DNA and maternal DNA and is

obtained from a sample from a pregnant female. The steps of the method of claim 85 include, among other steps, a step of incorporating nucleotides into digested DNA wherein (i) a nucleotide that terminates elongation, and is complementary to the locus of interest of an allele, is incorporated into the 5' overhang of the allele, and (ii) a nucleotide complementary to the locus of interest of a different allele is incorporated into the 5' overhang of the different allele, and the terminating nucleotide, which is complementary to a nucleotide in the 5' overhang of the different allele, is incorporated into the 5' overhang of the different allele. Claim 86 depends from independent claims 84 and 85.

As already noted, to establish a prima facie case of obviousness, the prior art references must teach or suggest all the claim limitations.

With respect to claims 84 and 86, as amended, Applicant submits that Jones et al., alone or in combination with Lo et al., neither teaches nor suggests all elements of claims 84 and 86. None of the cited references teaches or suggests the sequencing of template DNA that comprises a mixture of fetal DNA and maternal DNA. The methods of Lo et al. directed to the detection of chromosomal aberrations prenatally are focused on the detection and/or quantification of fetal DNA, not on sequencing reactions performed on template DNA comprising a mixture of fetal and maternal DNA. In particular, Lo et al. is focused on the use of paternally-derived sequences in fetal DNA that differ from the maternal DNA so as to selectively detect and amplify fetal DNA against the backdrop of maternal DNA. See, e.g., lines 3-26, page 5, line 27, page 31, to line 7, page 32, of Lo et al.

With respect to claims 85 and 86, as amended, Applicant submits that Jones et al., alone or in combination with Lo et al., neither teaches nor suggests all elements of the claims. Jones et al. neither teaches nor suggests the incorporation of a terminating nucleotide that is both complementary to the locus of interest of one allele and complementary to a nucleotide in the 5' overhang of a different allele. Jones et al. does not teach or suggest a method of determining a sequence of alleles at a locus of interest that uses the same terminating nucleotide in determining the sequence of one of the alleles as it does in determining the sequence of the other allele at the locus

of interest. Rather, Jones et al. describes methods for determining the sequence of polymorphisms using multiple, differentially labeled dideoxynucleotides for determining the sequence of the alleles at a locus of interest. See, e.g., paragraph [0067] on page 7, of Jones et al., which states that the “labeled probes can be two, three or four differentially labeled dideoxynucleotides,” but which fails to teach or suggest in any way that a single labeled dideoxynucleotide could suffice. See also paragraphs [0095] and [0097] on page 10, paragraph [0100] on page 11, and Figures 3B-3E of Jones et al., all of which illustrate the use of different labeled dideoxynucleotides in the Jones et al. method for distinguishing between the different alleles.

In addition, as also previously noted, to establish a prima facie case of obviousness, there must be some suggestion or motivation to modify the reference or combine reference teachings.

With respect to Claims 84-86, as amended, Applicant submits that one of ordinary skill in the art would not have been motivated to modify the teachings of Jones et al. by substituting a mixture of fetal DNA and maternal DNA obtained from a sample from a pregnant female for the DNA samples in the methods taught by Jones et al. Even if one of ordinary skill in the art was motivated by a desire to detect chromosomal aberrations prenatally, one would not have been motivated to apply the techniques taught by Jones et al. to a mixture of fetal DNA and maternal DNA. Neither Jones et al., nor Lo et al., teaches or suggests the desirability, feasibility, or utility of using a mixture of fetal and maternal DNA as template DNA, instead of fetal DNA as template DNA. Lo et al. is focused on methods of detecting chromosomal aberrations that are dependent upon selectively detecting and/or amplifying fetal DNA, as opposed to maternal DNA. The preference for amplifying and/or detecting fetal DNA, but not maternal DNA, is evident in the focus of the methods in Lo et al. which are directed to targeting paternally-derived sequences and/or sex-linked chromosome sequences found on the fetal DNA and not on the maternal DNA for amplification and/or detection. See, e.g., lines 3-26, page 5, line 27, page 31, to line 7, page 32. In effect, Lo et al. teaches away from the use of a mixture of fetal and maternal DNA in any form of prenatal chromosomal analysis.

Since Jones et al. in view of Lo et al., does not teach or suggest each and every element of claims 84-86, as amended, and since there is no motivation to combine the references, Applicant respectfully requests that the rejection of claims 84-86 be withdrawn.

Claims 143 and 145: The Office has rejected claims 143 and 145 as being unpatentable over Lapidus et al. (US 6,100,029) as applied against Claim 1 above and further in view of Shultz et al. (US 6,268, 146). Applicant respectfully traverses this rejection.

Claim 143 and 145 depend from independent claim 1, as amended. Accordingly, claims 143 and 145 are directed to a method for detecting a chromosomal abnormality, in which the template DNA comprises a mixture of fetal DNA and maternal DNA.

As already noted, to establish a prima facie case of obviousness, the prior art references must teach or suggest all the claim limitations. Applicant respectfully submits that Lapidus et al., alone or in combination with Shultz et al., does not teach or suggest all elements of claims 143 and 145. Lapidus et al., alone or in combination with Shultz et al., does not teach or suggest the use of template DNA comprising a mixture of fetal DNA and maternal DNA, as discussed above with respect to the rejection of claim 1 over Lapidus et al. under § 103.

Since Lapidus et al. in view of Jones et al. and further in view of Shultz et al. does not teach or suggest each and every element of claims 143 and 145, Applicant respectfully requests that the rejection of claim 143 and 145 under 35 USC § 103 be withdrawn.

Claims 144-146 and 148-149: The Office has rejected claims 144-146 and 148-149 as being unpatentable over Lo et al. (WO98/39474) in view of Wallace et al. (US 5,639,611) or Jones et al. (US 5,639,611) as applied against Claim 58 above and further in view of Shultz et al. (US 6,268, 146). Applicant respectfully traverses this rejection.

Claims 144-146 depend from independent claim 58 and therefore, like claim 58, are directed to methods comprising determining the sequence of a locus of interest on free fetal DNA from a sample comprising free fetal DNA, wherein an agent that inhibits cell lysis has been added to said sample to inhibit lysis of cells, if cells are present, wherein said agent is selected from the group

consisting of membrane stabilizer, cross-linker, and cell lysis inhibitor. Claims 148-149 have been amended to no longer depend from claim 144, although similar new claim 206 has been added. Claims 148-149, as amended, depend from independent claim 1 and therefore, like claim 1, as amended, are directed to methods for detecting the presence or absence of a fetal chromosomal abnormality, comprising quantitating the relative amount of the alleles at the heterozygous locus of interest, wherein said relative amount is expressed as a ratio, and wherein said ratio indicates the presence or absence of a fetal chromosomal abnormality, wherein the template DNA comprises a mixture of maternal and fetal DNA. In claims 148 and 149, an agent that inhibits cell lysis has been added to the sample to inhibit the lysis of cells, if present, wherein the agent is selected from the group consisting of membrane stabilizer, cross-linker, and cell lysis inhibitor.

As repeatedly noted above, to establish a prima facie case of obviousness, the prior art references must teach or suggest all the claim limitations. Applicant respectfully submits that the combination of Lo et al., in view of Wallace et al. or Jones et al., and further in view of Shultz et al. does not teach or suggest each and every element of claims 144-146 and 148-149. As discussed above with respect to the rejection of claim 58 and 87, Applicant submits that, contrary to the Office's unsupported assertions, Lo et al. does not teach or disclose the use of a cell lysis inhibitor, since EDTA is not a cell lysis inhibitor. In addition, with respect to claims 148-149, Applicant submits that the combination of Lo et al., in view of Wallace et al. or Jones et al., and further in view of Shultz et al., also fails to teach all elements of claims 148-149 because, for instance, the combination of references fails to teach quantitating and expressing as a ratio the relative amount of alleles at the heterozygous locus of interest where the template DNA comprising a mixture of maternal and fetal DNA.

Since Lo et al., in view of Wallace et al. or Jones et al., and further in view of Shultz et al. does not teach or suggest each and every element of claims 144-146 and 148-149, Applicant respectfully requests that the rejection of claims 144-146 and 148-149 under 35 USC 103 be withdrawn.

**Non-Statutory Obviousness-type Double Patenting Rejection**

The Office has rejected claims 84 and 86 under the judicially created doctrine of obviousness-type double patenting as being unpatentable over claims 1 and 5 of U.S. Patent No. 6,977,162. Applicant respectfully traverses this rejection.

As noted above, claim 84, as amended, is directed to a method for determining the sequence of a locus of interest from template DNA, wherein the template DNA comprises a mixture of fetal DNA and maternal DNA and is obtained from a sample from a pregnant female. Claim 86 depends from claim 84.

The only basis provided by the Office for the obviousness-type double patenting rejection of claims 84 and 86 over U.S. Patent No. 6,977,162 patent is that claim 84 “overlaps that of claim 1 of U.S. Patent No. 6,977,162.” Applicant respectfully submits that mere claim overlap is not the standard for obviousness-type double patenting rejections and that a rejection based on mere claim overlap is improper. As indicated in MPEP 804(II)(b)(1), “A double patenting rejection of the obviousness-type is ‘analogous to [a failure to meet] the nonobviousness requirement of 35 U.S.C. 103’ except that the patent principally underlying the double patenting rejection is not considered prior art.” A mere assertion of claim overlap is not sufficient to support a proper rejection under 35 USC § 103, and it does not support a proper obviousness-type double-patenting rejection.

Applicant submits that claims 84 and 86, as amended, are patentably distinct over claims 1 and 5 of U.S. Patent No. 6,977,162. Claims 84 and 86 are not obvious variations over claims 1 and 5 of U.S. Patent No. 6,977,162, since one of ordinary skill in the art, in the absence of Applicant’s teachings, would not have concluded, for instance, that the methods of claims 1 and 5 would be desirably, feasibly, and/or usefully applied to template DNA obtained from a sample from a pregnant female that comprises a mixture of fetal DNA and maternal DNA.

Since the rejection of claims 84 and 86 over U.S. Patent No. 6,977,162 is improper for the reasons stated above, Applicant respectfully requests that the rejection of claims 84 and 86 over

U.S. Patent No. 6,977,162 under the judicially created doctrine of obviousness-type double patenting be withdrawn.

The Office has also provisionally rejected claims 84 and 86 under the judicially created doctrine of obviousness-type double patenting as being unpatentable over Claims 1 and 4 of U.S. Patent Application 11/107,624.

Applicant notes that this is a provisional rejection only. Applicant will address this rejection, if maintained, at the appropriate time if conflicting claims are found allowable.

The Office has provisionally rejected claims 87 and 96-98 under the judicially created doctrine of obviousness-type double patenting as being unpatentable over Claim 1 of U.S. Patent Application 11/212,386.

Applicant notes that this is a provisional rejection only. Applicant will address this rejection, if maintained, at the appropriate time if conflicting claims are found allowable.

### **Claim Objections**

The Office has objected to claim 99 as being dependent upon a rejected independent base claim. Claim 99 is dependent upon rejected claims 98, 97, 96, and 87. Applicant submits that claims 87 and 96-98 are patentable over the cited references for the reasons indicated above. Since claim 99 is not dependent upon an unpatentable claim, Applicant respectfully requests that claim 99 be allowed.

### **Supplemental Information Disclosure Statement filed February 9, 2005**

A Supplemental Information Disclosure Statement and Form PTO/SB/08 were filed on February 9, 2005. A copy of the initialed Form PTO/SB/08 was not included with the Non-Final Office Action mailed March 17, 2006. Therefore, we respectfully request the Examiner to provide us with a copy of the initialed Form PTO/SB/08 for the Supplemental Information Disclosure Statement filed on February 9, 2005.

**CONCLUSION**

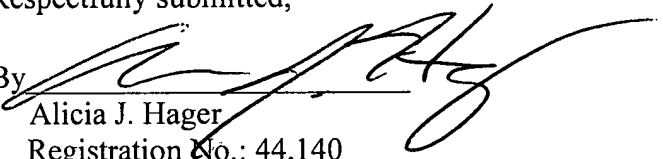
In view of the above, each of the presently pending claims in this application is believed to be in immediate condition for allowance. Accordingly, the Examiner is respectfully requested to withdraw the outstanding rejection of the claims and to pass this application to issue. If it is determined that a telephone conference would expedite the prosecution of this application, the Examiner is invited to telephone the undersigned at the number given below.

In the event the U.S. Patent and Trademark office determines that an extension and/or other relief is required, applicant petitions for any required relief including extensions of time and authorizes the Commissioner to charge the cost of such petitions and/or other fees due in connection with the filing of this document to **Deposit Account No. 03-1952** referencing docket no. **543312000420**. However, the Commissioner is not authorized to charge the cost of the issue fee to the Deposit Account.

Dated: July 14, 2006

Respectfully submitted,

By

  
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